

Best Available Copy

Naftali Primor, Philip Lazarovici  
Osborn Laboratories of Marine Science  
New York Aquarium  
Boardwalk & West 8th Street  
Brooklyn, NY 11224

ASSISTED BY A. Moran

20030117112

WORK UNIT NO. NR N00014-82-C-0435

CONTRACT

#### OBJECTIVES

Structure and mode of action of the shark repellent pardaxin(Px)

1984

DTIC  
ELECTE  
MAR 17 1985

#### ABSTRACT

A new column chromatography procedure, based on gel-permeation, ion exchange and chromatofocusing was employed to isolate the two main proteaceous, toxic, cytolytic and pore forming factors from the gland secretion of the Red Sea flatfish Pardachirus marmoratus. Pardaxin I(PXI) consisting 10% of the gland protein secretion, was shown to be 5-10 times more toxic, cytolytic and active on pore formation than pardaxin II (PXII) (8% of gland protein secretion). Gel-electrophoresis, amino acid analysis and N-terminal amino acid sequencing reveals a high degree of homogeneity and resemblance between the two toxins. They are rich in serine, glycine, alanine, leucine, and phenylalanine and devoid of arginine and tryptophan. Their N-terminal was found to be  $\text{NH}_2\text{-Gly-Phe-Phe}$ . Their hydrophobicity is evident from the chromatographic behavior on a hydrophobic resin, presence of nine successive hydrophobic residues on the  $\text{NH}_2$ -terminal, overall percentage

AD-A165 435

DISTRIBUTION STATEMENT A  
Approved for public release;  
Distribution Unlimited

DTIC FILE COPY

- 1 -

of toxins hydrophobicity and decreament of aqueous drops volume. The molecular weight of Pardaxin I in a monomer form is about 13 Kd as determined by gel filtration, gel electrophoresis and amino acid analyses. In aqueous solutions it tends to polymerize, mainly to a tetrameric form. Pardaxin I is composed of 120 amino acids and is free of carbohydrate and sialic acid residues. Mass spectrometry of the ethyl acetate extract of the gland secretion and the purified toxins reveals the presence of sterols in the secretion but their absence in the purified toxins.

Pardaxin I was iodinated without affecting its chemical and pore forming properties. It binds to liposomes, of different phospholipid compositions. In hyperpolarized unilamellar liposomes and multilamellar liposomes with an imposed chemical gradient, pardaxin I produced a fast, non,ion-specific ( $10^{-10}M$ ) and slow, cation-specific pore ( $10^{-8}M$ ) respectively (in preparation).

We have shown that in fish the gill tissue is the main organ for the action of PX. This action is being correlated with the channel-forming activity of PX. Our findings indicate that PX displays a channel or pore activity, e.g., it produces an increase in conductance in an artificial lipid membrane and in liposomes. The interaction of PX with fish gill tissue is being investigated presently.

-2-



by <i>ltr on file</i>	
Distribution /	
Availability Codes	
Dist	Avail and/or Special
A-1	

Moran, A., Korchak, Z., Moran, N. and Primor, N.  
Surfactant and channel-forming activities of the Moses Sole  
toxin. In, Toxins, drugs, and pollutants in marine animals.  
eds., L. Bolis, J. Zadunaisky and R. Gilles.  
Springer-Verlag Berlin, Heidelberg, New York, Tokyo. 1984  
p. 13-15.

N. Primor. Pardaxin produces postjunctional muscle  
contraction in guinea-pig intestinal smooth muscle. Br. J.  
Pharmacol. 82, 43-49 (1984).

N. Primor, Pharyngeal cavity and the gills are the  
target organ for the repellent action of pardaxin in shark.  
Experientia. 41, 693-695 (1985).